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Please find below and/or attached an Office communication concerning this application or proceeding.

| Office Action Summary | Application No. | Applicant(s) | |
|---|---|---|---|
| | 10/014,326 | JAKOBSEN ET AL. | |
| | Examiner | Art Unit | |
| | MINH-TAM DAVIS | 1642 | |

*-- The MAILING DATE of this communication appears on the cover sheet with the correspondence address --*

**Period for Reply**

A SHORTENED STATUTORY PERIOD FOR REPLY IS SET TO EXPIRE 3 MONTH(S) FROM THE MAILING DATE OF THIS COMMUNICATION.

- Extensions of time may be available under the provisions of 37 CFR 1.136(a). In no event, however, may a reply be timely filed after SIX (6) MONTHS from the mailing date of this communication.
- If the period for reply specified above is less than thirty (30) days, a reply within the statutory minimum of thirty (30) days will be considered timely.
- If NO period for reply is specified above, the maximum statutory period will apply and will expire SIX (6) MONTHS from the mailing date of this communication.
- Failure to reply within the set or extended period for reply will, by statute, cause the application to become ABANDONED (35 U.S.C. § 133). Any reply received by the Office later than three months after the mailing date of this communication, even if timely filed, may reduce any earned patent term adjustment. See 37 CFR 1.704(b).

**Status**

1)☒ Responsive to communication(s) filed on 24 November 2003.

2a)☐ This action is **FINAL**. 2b)☒ This action is non-final.

3)☐ Since this application is in condition for allowance except for formal matters, prosecution as to the merits is closed in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11, 453 O.G. 213.

**Disposition of Claims**

4)☒ Claim(s) 22-34 is/are pending in the application.

4a) Of the above claim(s) _____ is/are withdrawn from consideration.

5)☐ Claim(s) _____ is/are allowed.

6)☒ Claim(s) 22-34 is/are rejected.

7)☐ Claim(s) _____ is/are objected to.

8)☐ Claim(s) _____ are subject to restriction and/or election requirement.

**Application Papers**

9)☐ The specification is objected to by the Examiner.

10)☐ The drawing(s) filed on _____ is/are: a)☐ accepted or b)☐ objected to by the Examiner.

Applicant may not request that any objection to the drawing(s) be held in abeyance. See 37 CFR 1.85(a).

Replacement drawing sheet(s) including the correction is required if the drawing(s) is objected to. See 37 CFR 1.121(d).

11)☐ The oath or declaration is objected to by the Examiner. Note the attached Office Action or form PTO-152.

**Priority under 35 U.S.C. § 119**

12)☐ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).

a)☐ All b)☐ Some * c)☐ None of:

1.☐ Certified copies of the priority documents have been received.

2.☐ Certified copies of the priority documents have been received in Application No. _____.

3.☐ Copies of the certified copies of the priority documents have been received in this National Stage application from the International Bureau (PCT Rule 17.2(a)).

* See the attached detailed Office action for a list of the certified copies not received.

**Attachment(s)**

1) ☒ Notice of References Cited (PTO-892)

2) ☐ Notice of Draftsperson's Patent Drawing Review (PTO-948)

3) ☒ Information Disclosure Statement(s) (PTO-1449 or PTO/SB/08) Paper No(s)/Mail Date 03/25/02.

4) ☐ Interview Summary (PTO-413) Paper No(s)/Mail Date. _____ .

5) ☐ Notice of Informal Patent Application (PTO-152)

6) ☐ Other: _____.

## DETAILED ACTION

Applicant's election with traverse of group I, claims 22-34 in Paper of 10/28/03 is acknowledged and entered.

Claims 22-34 are pending in the instant application.

**Claims 22-34 are examined in the instant application**, wherein claims 22-34 are examined only to the extent of a soluble T cell receptor comprising alpha and beta chains.

The traverse is as follows:

The $\alpha$ and $\gamma$ chains are highly related structurally and functionally and evolutionary. Similarly, the $\beta$ and $\delta$ chains are highly related structurally and functionally and evolutionary. Applicant therefore, requests that the restriction requirement be modified to a requirement for election of species, and that the Examiner first search and examine the elected species of alpha and beta chains.

The arguments are not found to be persuasive for the following reasons:

The $\alpha$-$\beta$ and $\gamma$-$\delta$ forms, although having some similarity in structure, have quite distinct anatomical locations and probably function, as disclosed in the specification (p.3, lines 30-31). Therefore, the searches for different TCR forms are not co-extensive, and it would be a serious burden for the Examiner to search all the groups together.

Accordingly the restriction requirement was and is still deemed to be proper, and therefore made FINAL.

**OBJECTION**

Claims 22-34 are objected to because part of claims 22-34 are drawn to non-elected invention, i.e. a soluble T cell receptor comprising $\gamma$ and $\delta$ chains.

**REJECTION UNDER 35 USC 112, SECOND PARAGRAPH**

Claim 23 is rejected under 35 U.S.C. 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention.

Claim 23 is indefinite for the use of the language "low concentration". The term "low concentration" in claim 32 is a relative term which renders the claim indefinite. The term "low concentration" is not defined by the claim, the specification does not provide a standard for ascertaining the requisite degree, and one of ordinary skill in the art would not be reasonably apprised of the scope of the invention.

**REJECTION UNDER 35 USC 112, FIRST PARAGRAPH, NEW MATTER**

Claim 24 is rejected under 35 U.S.C. 112, first paragraph, as the specification does not contain a written description of the claimed invention. .

Claim 24 is drawn to a recombinant TCR according to claim 22, wherein said TCR is stable at a concentration of about "20 mg/ml".

The limitation of a recombinant TCR which is stable at a concentration of about "20 mg/ml" has no clear support in the specification and the claims as originally filed.

A review of the specification shows support for stability of the claimed TCR at relatively low concentration, at concentrations below 1mg/ml and preferably about 10

ug/ml (p.9, lines 27-29). The specification also shows support for specific binding of the TCR zipper obtained a low concentrations (at least 40 ug/ml), implying that the TCR zipper is relatively stable (p. 43, line12-14).

**The subject matter claimed in claim 24 broadens the scope of the invention as originally disclosed in the specification.**

**REJECTION UNDER 35 USC 112, FIRST PARAGRAPH, SCOPE**

Claim 33 is rejected under 35 U.S.C. 112, first paragraph, because the specification, while being enabling for a soluble T cell receptor (TCR), comprising a TCR alpha and a beta extracellular domain having at C-terminal, a first and a second heterologous dimerization peptides, respectively, wherein the dimerization domain are specifically heterodimerized to form a heterodimerisation domain, wherein a disulphide bond present in native TCRs between the alpha and beta chain is absent, **does not reasonably provide enablement for a soluble T cell receptor (TCR) linked to a therapeutic agent,** wherein said TCR comprises a TCR alpha and beta extracellular domain, having at C-terminal a first and a second heterologous dimerization peptides, respectively, wherein the dimerization domains are specifically heterodimerized to form a heterodimerisation domain, and wherein a disulphide bond present in native TCRs between the alpha and beta chain is absent. The specification does not enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to make and use the invention commensurate in scope with these claims.

Claim 33 is drawn to a recombinant soluble T cell receptor (TCR) linked to a therapeutic agent, wherein said TCR comprises a TCR alpha and beta extracellular domain, each having a C-terminal dimerization peptide, wherein the dimerization domains are specifically heterodimerized to form a heterodimerisation domain, and wherein a disulphide bond present in native TCRs between the alpha and beta chain is absent

The specification discloses that specific binding of TCR-zipper to viral peptide-MHC is obtained even at low concentration, at least 40 ug/ml, implying that the TCR zipper is relatively stable (Example 2 on pages 42-43, especially p.43, second paragraph). The specification contemplates linking a therapeutic agent such as a toxic moiety to the claimed recombinant TCR, such as TCR specific for tumor antigens, for use in cell killing, such as tumor cell (p.15).

Claim 33 encompasses a recombinant soluble T cell receptor (TCR) linked to a therapeutic agent for use in therapy such as treating cancer, as contemplated.

One cannot extrapolate the teaching in the specification to the scope of the claim, because it is unpredictable that the claimed soluble TCR linked to a therapeutic agent would be effective and thus could be used for therapy, as contemplated, in view that it is well known that the art of anticancer drug discovery for cancer therapy is highly unpredictable, for example, Gura (Science, 1997, 278:1041-1042) teaches that researchers face the problem of sifting through potential anticancer agents to find ones promising enough to make human clinical trials worthwhile and teach that since formal screening began in 1955, many thousands of drugs have shown activity in either cell or

animal models but that only 39 have actually been shown to be useful for chemotherapy (p. 1041, see first and second para). Because of the known unpredictability of the art, in the absence of experimental evidence, no one skilled in the art would accept the assertion that the claimed peptide would be useful for treating cancer. Further, the refractory nature of cancer to drugs is well known in the art. Jain (Sci. Am., 1994, 271:58-65) teaches that tumors resist penetration by drugs (p.58, col 1) and that scientists need to put expanded effort into uncovering the reasons why therapeutic agents that show encouraging promise in the laboratory often turn out to be ineffective in the treatment of common solid tumors (p. 65, col 3). Curti (Crit. Rev. in Oncology/Hematology, 1993, 14:29-39) teaches that solid tumors resist destruction by chemotherapy agents and that although strategies to overcome defense mechanisms of neoplastic cells have been developed and tested in a number of patients, success has been limited and further teaches that it is certainly possible that cancer cells possess many as yet undefined additional molecular mechanisms to defeat chemotherapy treatment strategies and if this is true, designing effective chemotherapeutic regimens for solid tumors may prove a daunting task (para bridging pages 29-30) and concludes that knowledge about the physical barriers to drug delivery in tumors is a work in progress (p. 36, col 2). It is clear that based on the state of the art, in the absence of experimental evidence, no one skilled in the art would accept the assertion that the claimed peptide would be useful for treating cancer. In addition, Hartwell et al (Science, 1997, 278:1064-1068) teach that an effective chemotherapeutic must selectively kill tumor cells, that most anticancer drugs have been discovered by serendipity and that

the molecular alterations that provide selective tumor cell killing are unknown and that even understanding the detailed molecular mechanism by which a drug acts often provides little insight into why the treated tumor cell dies (para bridging pages 1064-1065) and Jain (cited supra) specifically teaches that systemic treatment typically consists of chemotherapeutic drugs that are toxic to dividing cells (p. 58, col 2, para 2).

Further, one cannot predict that the claimed soluble TCR linked to a therapeutic agent would be effective for possible therapy for cancer and diseases involving immune system regulation, such as allergies, autoimmune diseases, infections, and transplant rejection. A therapeutic agent must accomplish several tasks to be effective. It must be delivered into the circulation that supplies the target cells and interact at the proper site of action and must do so at a sufficient concentration and for a sufficient period of time. Further, variables such as biological stability, half-life or clearance from the blood are important parameters in achieving successful therapy. The soluble TCR may be inactivated *in vivo* before producing a sufficient effect, for example, by proteolytic degradation, immunological activation or due to an inherently short half life of the protein and the *in vitro* tests of record do not sufficiently duplicate the conditions which occur *in vivo*. In addition, the soluble TCR may not otherwise reach the target because of its inability to penetrate tissues or cells where its activity is to be exerted, may be absorbed by fluids, cells and tissues where the soluble TCR has no effect, circulation into the target area may be insufficient to carry the soluble TCR and a large enough local concentration may not be established.

Thus, in view of the teaching in the art, one cannot predict that the claimed soluble TCR linked to a therapeutic agent would be effective for therapy, as contemplated, and one would not know how to use the claimed invention.

In view of the above, it would be undue experimentation for one of skill in the art to practice the claimed invention.

**REJECTION UNDER 35 USC 102(b)**

The following is a quotation of the appropriate paragraphs of 35 U.S.C. 102 that form the basis for the rejections under this section made in this Office action:

> A person shall be entitled to a patent unless –
>
> (b) the invention was patented or described in a printed publication in this or a foreign country or in public use or on sale in this country, more than one year prior to the date of application for patent in the United States.

**1.** Claims 22-26, 34 are rejected under 35 USC 102(b) as being anticipated by Gregoire, C et al, 1991, PNAS, USA, 88: 8077-8081.

Claims 22-26 are drawn to a recombinant soluble T cell receptor (TCR), comprising a TCR alpha and beta extracellular domain, each having a C-terminal dimerization peptide, wherein the dimerization domains are specifically heterodimerized to form a heterodimerisation domain, and wherein a disulphide bond present in native TCRs between the alpha and beta chain is absent (claim 22). Said recombinant TCR is

stable at low concentrations, or at about 10 ug/ml, 20 mg/ml, or at concentration below 1 mg/ml (claims 23-26).

Claim 34 is drawn to a recombinant non-membrane-bound T cell receptor produced by expressing said recombinant soluble T cell receptor of claim 22.

Claim 34 is drawn to a product by process, and thus is treated as the product per se.

Gregoire, C et al teach that the extracellular V and C domains of the TCR alpha and beta chains have been previously linked to the lipid linker (p.8077, second column, third line). Thus from the teaching of Gregoire, C et al, it is clear that the extracellular domain of TCR comprises V and C domains of the TCR alpha and beta chains.

Further, Gregoire, C et al teach chimeric soluble T cell receptor alpha beta heterodimers linked to immunoglobulin constant Ck region (p.8078, second column, last paragraph, figure 1 on page 8078, and figure 4 on page 8080). Gregoire, C et al teach that the alpha chain consists of the TCR V alpha region joined to the first exon of TCR C alpha (C alpha 1), and the C region of the immunogloblin k L chain (Ck), and the beta chain consists of the TCR V beta region joined to the first exon of TCR C beta (C beta 1), and the immunoglobulin Ck (p.8078, second column, last paragraph, figure 1 on page 8078, and figure 4 on page 8080).

Gregoire, C et al teach that the chimeric polypeptides do not include the second exon of the TCR C alpha and C beta gene, and therefore lack the cysteine residues normally involved in the covalent linkage of the TCR alpha and beta chains (p.8080, first column, lines 2-6). Gregoire, C et al teach that the abililty of their construction to form

alpha-k-beta-k dimers may be due the lack of the cysteine residue located at the COOH- terminal to the end of the C alpha and C beta regions (p.8080, second column, first paragraph).

Gregoire, C et al teach that an illegitimate interaction between the COOH terminus of a TCR C domain and the NH2-termus of a Ck domain may distort the proper pairing of the Ck domains and accordingly their ability to be disulfide linked (p. 8080, first column, last 8 lines bridging second column). In other words, the Ck domains would pair with each other, or dimerize.

Thus the chimeric soluble TCR taught by Gregoire, C et al seems to be the same as the claimed soluble recombinant TCR.

Although the reference does not teach that the soluble TCR is stable at low concentration, or at a concentration of about 10 ug/ml or 20 mg/ml, or at a concentration below 1 mg/ml, however, the claimed soluble TCR appears to be the same as the prior art soluble TCR. The office does not have the facilities and resources to provide the factual evidence needed in order to establish that the product of the prior art does not possess the same material, structural and functional characteristics of the claimed product. In the absence of evidence to the contrary, the burden is on the applicant to prove that the claimed product is different from those taught by the prior art and to establish patentable diffrences. See In re Best 562F.2d 1252, 195 USPQ 430 (CCPA 1977) and Ex parte Gray 10 USPQ 2d 1922 (PTO Bd. Pat. App. & Int. 1989).

**2.** Claims 22-26, 34 are rejected under 35 USC 102(b) as being anticipated by Weber, S et al, 1992, Nature, 356 (6372): 793-6, IDS of 03/25/02.

Claims 22-26 are drawn to a recombinant soluble T cell receptor (TCR), comprising a TCR alpha and beta extracellular domain, each having a C-terminal dimerization peptide, wherein the dimerization domains are specifically heterodimerized to form a heterodimerisation domain, and wherein a disulphide bond present in native TCRs between the alpha and beta chain is absent (claim 22). Said recombinant TCR is stable at low concentrations, or at about 10 ug/ml, 20 mg/ml, or at concentration below 1 mg/ml (claims 23-26).

Claim 34 is drawn to a recombinant non-membrane-bound T cell receptor produced by expressing said recombinant soluble T cell receptor of claim 22.

Claim 34 is drawn to a product by process, and thus is treated as the product per se.

Weber et al teach soluble TCR-immunoglobulin chimera, constructed with the alpha- and beta- chains linked to the immunoglobulin light chain constant region (Ck). The alpha- and beta- chains comprise the TCR alpha and beta variable region and the first constant region exons (p.793, second column, paragraph before last, and figure 1 on page 794). Weber et al teach soluble TCR-immunoglobulin chimera is biologically active: it specifically inhibits antigen-dependent activation of the relevant T-cell clone and thus discriminates between proper and irrelevant peptides presented by major histocompatibility complex molecules (abstract and figure 3 on page 795).

It is noted that the TCR alpha and beta variable (V) and constant (C) region of the TCR chimera taught by Weber et al is an extracellular domain, because Gregoire, C et al,1991, PNAS, USA, 88: 8077-8081, teach that the "extracellular" V and C domains

of the TCR alpha and beta chains have been previously linked to the lipid linker (p.8077, second column, third line), thus indicating that the V and C domains of the TCRs are extracellular domain.

In addition, it is noted that the first exon of the TCR C alpha and C beta gene does not have the cysteine residues normally involved in the covalent linkage between the TCR alpha and beta chains because Gregoire, C et al teach chimeric TCR alpha-beta polypeptides, which is linked to immunoglobulin Ck, wherein said polypeptides contain the first exon of the TCR C alpha and C beta gene, but do not include the second exon of the TCR C alpha and C beta gene, and therefore lack the cysteine residues normally involved in the covalent linkage of the TCR alpha and beta chains (p.8080, first column, lines 2-6).

Further, it is noted that the Ck domains taught by Weber et al would pair with each other, or dimerize, because Gregoire, C et al teach that an illegitimate interaction between the COOH terminus of a TCR C domain and the NH2-termus of a Ck domain may distort the proper pairing of the Ck domains and accordingly their ability to be disulfide linked (p. 8080, first column, last 8 lines bridging second column).

Thus, the soluble TCR taught by Weber et al seems to be the same as the claimed soluble TCR.

Although the reference does not teach that 1) the TCR V and C domains of the soluble TCR chimera is the TCR extracellular domain, 2) a disulphide bond present in native TCRs between the alpha and beta chain is absent in said TCR chimera and 3) the soluble TCR chimera is stable at low concentration, or at a concentration of about

10 ug/ml or 20 mg/ml, or at a concentration below 1 mg/ml, however, the claimed soluble TCR appears to be the same as the prior art soluble TCR. The office does not have the facilities and resources to provide the factual evidence needed in order to establish that the product of the prior art does not possess the same material, structural and functional characteristics of the claimed product. In the absence of evidence to the contrary, the burden is on the applicant to prove that the claimed product is different from those taught by the prior art and to establish patentable diffrences. See In re Best 562F.2d 1252, 195 USPQ 430 (CCPA 1977) and Ex parte Gray 10 USPQ 2d 1922 (PTO Bd. Pat. App. & Int. 1989).

**REJECTION UNDER 35 USC 103**

The following is a quotation of 35 U.S.C. 103(a) which forms the basis for all obviousness rejections set forth in this Office action:

> (a) A patent may not be obtained though the invention is not identically disclosed or described as set forth in section 102 of this title, if the differences between the subject matter sought to be patented and the prior art are such that the subject matter as a whole would have been obvious at the time the invention was made to a person having ordinary skill in the art to which said subject matter pertains. Patentability shall not be negatived by the manner in which the invention was made.

The factual inquiries set forth in *Graham* **v.** *John Deere Co.*, 383 U.S. 1, 148 USPQ 459 (1966), that are applied for establishing a background for determining obviousness under 35 U.S.C. 103(a) are summarized as follows:

1. Determining the scope and contents of the prior art.
2. Ascertaining the differences between the prior art and the claims at issue.
3. Resolving the level of ordinary skill in the pertinent art.
4. Considering objective evidence present in the application indicating obviousness or nonobviousness.

This application currently names joint inventors. In considering patentability of the claims under 35 U.S.C. 103(a), the examiner presumes that the subject matter of the various claims was commonly owned at the time any inventions covered therein were made absent any evidence to the contrary. Applicant is advised of the obligation under 37 CFR 1.56 to point out the inventor and invention dates of each claim that was not commonly owned at the time a later invention was made in order for the examiner to consider the applicability of 35 U.S.C. 103(c) and potential 35 U.S.C. 102(e), (f) or (g) prior art under 35 U.S.C. 103(a).

**1.** Claims 22-27, 29, 34 are rejected under 35 USC 103(a) as being obvious over Chang et al, 1994, Proc Natl Acad Sci, USA, 91: 11408-11412, IDS of 03/25/02, in view of Gregoire et al, 1991, supra, Garboczi et al, 1996, J Immunol, 157(12): 5403-10, IDS of 03/25/02, and Wulfing et al, 1994, J Mol Biol, 242: 655-669.

Claims 22-27, 29 are drawn to a recombinant soluble T cell receptor (TCR), comprising a TCR alpha and beta extracellular domain, each having a C-terminal

dimerization peptide, wherein the dimerization domains are specifically heterodimerized to form a heterodimerisation domain, and wherein a disulphide bond present in native TCRs between the alpha and beta chain is absent (claim 22). Said recombinant TCR is stable at low concentrations, or at about 10 ug/ml, 20 mg/ml, or at concentration below 1 mg/ml (claims 23-26). The heterodimerisation domain is a coiled coil domain (claim 27). Said recombinant TCR comprises a flexible linker located between the TCR chains and the dimerization peptides (claim 29).

Claim 34 is drawn to a recombinant non-membrane-bound T cell receptor produced by expressing said recombinant soluble T cell receptor of claim 22.

Claim 34 is drawn to a product by process, and thus is treated as the product per se.

It is noted that since there is no limitation that the TCR extracellular domain is a full length TCR extracellular domain, one could reasonably interpret that the TCR extracellular domain could comprise either the variable (V) domain, or both the variable (V) and constant (C) domains.

Chang et al teach a soluble TCR, in which the TCR alpha and beta V and C extracellular segments are fused at the carboxyl terminal to 30 amino acid long segments via a cleavable flexible linker (abstract and figure 2 on page 11410). Chang et al teach that the peptide segments have been previously shown to selectively associate to form a stable heterodimeric coiled coil, termed leucine zipper (abstract, and p.11408, second column, first three lines of second paragraph). Chang et al teach that the disulfide-linked alpha beta heterodimer is the clonally unique component that possesses

a recognitition site for antigen in the context of the major histocompatibility complex (p. 11408, first column, first paragragph after abstract). Chang et al teach that the heterodimeric TCR protein is produced from baculovirus-infected high 5 cells (figure 3 legend on page 11411). Chang et al teach that the structure of the heterodimeric TCR protein is judged to be native when probed with a panel of 17 monoclonal antibodies specific for the alpha and beta constant and variable domains (abstract, and p.11411, last paragraph bridging p.11412). Chang et al further teach that now it should be possible to facilitate association of any type of naturally occurring heterodimeric structure, and that association between individual protein domains, such as TCR V alpha and V beta can be fostered in the absence of other protein segments (C alpha and C beta) (p.11412, second column).

Chang et al do not teach that a disulphide bond present in native TCRs between the alpha and beta chain is absent in the recombinant TCR having both the alpha and beta variable and constant domains. Chang et al do not teach that said recombinant TCR is stable at low concentrations, or at about 10 ug/ml, 20 mg/ml, or at concentration below 1 mg/ml .

Gregoire, C et al teach chimeric soluble T cell receptor alpha beta heterodimers linked to immunoglobulin constant Ck region (p.8078, second column, last paragraph, figure 1 on page 8078, and figure 4 on page 8080). Gregoire, C et al teach that the alpha chain consists of the TCR V alpha region joined to the first exon of TCR C alpha (C alpha 1), and the C region of the immunogloblin k L chain (Ck), and the beta chain consists of the TCR V beta region joined to the first exon of TCR C beta (C beta 1), and

the immunoglobulin Ck (p.8078, second column, last paragraph, figure 1 on page 8078, and figure 4 on page 8080). Gregoire, C et al teach that the chimeric polypeptides do not include the second exon of the TCR C alpha and C beta gene, and therefore lack the cysteine residues normally involved in the covalent linkage of the TCR alpha and beta chains (p.8080, first column, lines 2-6).

Gregoire, C et al teach that in C-type pairing the NH2 termini of the TCR C regions are far apart, whereas the COOH termini are closed to each other and constrained by a disulfide bond, and that in both of their chimeric chains, the COOH terminus of a TCR C domain has to be fitted onto the NH2 terminus of an immunoglobulin Ck domain (p.8080, first column, last 8 lines). Gregoire, C et al teach that such an illegitimate interaction may distort the proper pairing of the Ck domains and accordingly their ability to be disulfide linked ((p.8080, first column, last three lines, bridging second column) . Gregoire, C et al teach that with regard to other, unsuccessful attempts, it should be noted that the unique ability of their construction to form alpha-k-beta-k dimers may be due the lack of the cysteine residue located at the COOH-terminal to the end of the C alpha and C beta regions (p.8080, second column, first paragraph).

It is clear from figure 2 on page 11410, in the reference by Chang et al, and from the teaching of Gregoire, C et al that without the C domains, a conjugate of TCR V alpha and V beta taught by Chang et al would not have the interchain disulfide bond, because the interchain disulfide is not found in the TCR V alpha and V beta.

Further, from the teaching of Gregoire, C et al one of ordinary skill in the art would have expected that **the lack of the disulfide bond located at the COOH-terminal to the end of the C alpha and C beta regions facilitates the pairing of the dimerizing immunoglobulin constant region Ck.**

Garboczi et al teach expression of the extracellular domains of the TCR alpha and beta chains separately as insoluble inclusion bodies in *E. coli* (p.5404, first column, second paragragph). Garboczi et al teach that refolded and mixed, the alpha and beta chains form soluble heterodimers, whether linked by an interchain disulfide bond or not. Garboczi et al teach that heterodimerization and antigenic specificity of TCR do not require its interchain disulfide bond, transmembrane segments or glycosylation (abstract). Garboczi et al teach that the alpha and beta chains of TCR expressed without the interchain disulfide bonds, when refolded together, form heterodimers spontaneously and at higher yield than the alpha and beta chains of TCR expressed with the interchain disulfide bonds (emphasis added) (p.5404, first column, second paragraph). Garboczi et al teach that the refolded noncovalently associated TCR is stable and very soluble and is routinely prepared at 100mg/ml (2 mM), and binds specifically to the ligand HLA-A2/Tax (p.5406, second column, last two paragraph, and p.5407, first column, paragraph before last).

Wulfing et al teach that T-cell receptor fragments are correctly folded in the periplasm of E. coli. Wulfing et al teach that the correctly folded scTCR is essentially stable, whereas misfolded scTCR is rapidly degraded, and that the effect of increased yields in the direct over-expression system is due to enhanced in vivo folding, working

synergistically with substantially higher intrinsic proteolytic stability of the correctly folded material (p.665, second column, last two paragraph, bridging p. 666).

From the teaching of Wulfing et al one would have expected that higher yields of TCRs are correlated with correct TCR folding, and thus **a decrease in the yield of alpha and beta chains of TCR expressed with the interchain disulfide bonds,** as compared to alpha and beta chains of TCR expressed without the interchain disulfide bonds as taught by Garboczi et al, **indicates an increase in misfolding** of alpha and beta chains of TCR expressed **with the interchain disulfide bonds.** In other words, the interchain disulfide bond seems to interfere with the folding of the alpha and beta chains of TCRs.

Thus from the teaching of Garboczi et al and Wulfing et al, one of ordinary skill in the art would expect that the interchain disulfide bond of TCR not only is not necessarily for TCR binding to the target peptide, but also could interfere with the folding of the alpha and beta chains, resulting in lower yield.

Therefore, it would have been prima facia obvious to one of ordinary skill in the art at the time the invention was made to combine the teaching of Chang et al, Gregoire, C et al and Garboczi et al, Wulfing et al for making a chimeric TCR taught by Chang et al, wherein the interchain disulfide bond between the alpha and beta chains is omitted, because of the following reasons:

1) One of ordinary skill in the art would have expected that the lack of the disulfide bond located at the COOH- terminal to the end of the C alpha and C beta

regions would facilitate the paring of the dimerizing peptide, in view of the teaching by Gregoire, C et al,

2) One of ordinary skill in the art would have expected that the interchain disulfide bond of TCR not only is not necessarily for its binding to the target peptide, but also could interfere with the folding of the alpha and beta chains, resulting in lower yield, in view of the teaching of Garboczi et al, and Wulfing et al, and

3) Chang et al teach that association between individual protein domains, such as TCR V alpha and V beta can be fostered in the absence of other protein segments (C alpha and C beta).

In other words, one of ordinary skill in the art would have been motivated to omit the interchain disulfide bond of the chimeric TCR, to further improve the dimerization and the yield of the chimeric TCR, in view of the teaching of Gregoire, C et al, Garboczi et al, and Wulfing et al, and because Chang et al teach that association between individual protein domains, such as TCR V alpha and V beta can be fostered in the absence of other protein segments (C alpha and C beta).

One of ordinary skill in the art would have been motivated to omit the interchain disulfide bond of the chimeric TCR taught by Chang et al with a reasonable expectation of success.

**2.** Claim 28 is rejected under 35 U.S.C. 103(a) as being unpatentable over Chang et al, 1994, supra, in view of Gregoire et al, 1991, supra, Garboczi et al, 1996, J Immunol, 157(12): 5403-10, IDS of 03/25/02, and and Wulfing et al, 1994, J Mol Biol,

242: 655-669 as applied to claims 22-27, 29, 34 above, and further in view of US 5,643,731 or US 5,582,996.

Claim 28 is drawn to the recombinant of claim 27, wherein the dimerisation peptides are c-jun and v-fos dimerisation peptide (claim 28).

The teaching of Chang et al, Gregoire et al, Garboczi et al, and Wulfing et al has been set forth above.

Chang et al, Gregoire et al, Garboczi et al, and Wulfing et al do not teach that the dimerisation peptides are c-jun and v-fos dimerisation peptides.

US 5,643,731 teach that c-jun and v-fos are particularly the preferred leucine zipper peptides (column 3, lines 38-42). US 5,643,731 teach that c-jun and v-fos leucine zipper peptides do not homodimerize, and together form very stable heterodimeric complexes (column 3, lines 26-37).

US 5,582,996 teach that the dimeric Fab is formed when a Fab with a c-fos leucine zipper is mixed with a Fab with a c-jun leucine zipper. US 5,582,996 teach that the hinge region of the dimeric Fab retains sufficient flexibility to permit the Fab fragments to bind antigen in a manner similar to that of normal antibodies (Example 7, on columns 15-16).

It would have been obvious to one of ordinary skill in the art at the time the invention was made to combine the teaching of Chang et al, Gregoire et al, Garboczi et al, and Wulfing et al with the teaching of US 5,643,731 or US 5,582,996, and use the c-jun and v-fos dimerisation peptides as the leucine zipper dimerization peptides taught by Chang et al, because c-jun and v-fos dimerisation peptides are preferred leucine

zipper peptides, which do not homodimerize, and together form very stable heterodimeric complexes, as taught by US 5,643,731, and because c-jun and v-fos leucine zipper peptides are commonly used to dimerize antigen binding fragments, such as Fab fragment, without as taught by US 5,582,996.

**3.** Claim 30 is rejected under 35 U.S.C. 103(a) as being unpatentable over Chang et al, 1994, supra, in view of Gregoire et al, 1991, supra, Garboczi et al, 1996, J Immunol, 157(12): 5403-10, IDS of 03/25/02, and and Wulfing et al, 1994, J Mol Biol, 242: 655-669 as applied to claims 22-27, 29, 34 above, and further in view of Arcone, R, 1991, Eur J biochem, 198(3): 541-7.

Claim 30 is drawn to the recombinant of claim 22, wherein the recombinant TCR is expressed in an E. coli expression system (claim 30).

The teaching of Chang et al, Gregoire et al, Garboczi et al, and Wulfing et al has been set forth above.

Although Chang et al do not teach expression of the TCR complex in an E. coli expression system, Garboczi et al teach expression of the extracellular domains of the TCR alpha and beta chains separately as insoluble inclusion bodies in *E. coli,* and Wulfing et al teach that T-cell receptor fragments are correctly folded in the periplasm of E. coli.

Acorne et al teach that expression in E coli of a human interleukin-6 has the advantage of producing a high yield.

It would have been obvious to one of ordinary skill in the art at the time the invention was made to express in E. coli the soluble TCR without interchain disulfide

bond, taught by the combination of the teaching of Chang et al, Gregoire et al, Garboczi et al, and Wulfing et, using the method taught by Garboczi et al, or Wulfing et, because the E coli expression system provides a high yield of the recombinant protein, as taught by Acorne et al, and because it is well known in the art that E. coli is easy to handle and grow quickly in large quantity.

**4.** Claims 31-32 are rejected under 35 U.S.C. 103(a) as being unpatentable over Chang et al, 1994, supra, in view of Gregoire et al, 1991, supra, Garboczi et al, 1996, J Immunol, 157(12): 5403-10, IDS of 03/25/02, and Wulfing et al, 1994, J Mol Biol, 242: 655-669 as applied to claims 22-27, 29, 34 above, and further in view of US 5,635,363.

Claims 31-32 are drawn to the recombinant of claim 22, which is biotinylated at the C-terminus or is labeled with a detectable label.

The teaching of Chang et al, Gregoire et al, Garboczi et al, and Wulfing et al has been set forth above.

In addition, Chang et al teach that a soluble TCR, in which the alpha and beta TCR extracellular segments are fused at the carboxyl terminal to 30 amino acid long dimerizing segments via a cleavable flexible linker (abstract and figure 2 on page 11410). Thus, one would have expected that the C-terminus of the soluble TCR would be the C-terminus end of the dimerizing peptide.

Moreover, Chang et al teach that the disulfide-linked alpha beta heterodimer is the clonally unique component that possesses a recognitition site for antigen in the context of the major histocompatibility complex. Thus, one would have expected that the

N-terminal region is where the alpha and beta chains are, and where the recognition site for antigen is.

Chang et al, Gregoire et al, Garboczi et al, and Wulfing et al do not teach that the soluble TCR is biotinylated at the C-terminus and labeled with a detectable label.

US 5,635,363 teaches biotinylation and labeling of a complex of alpha and beta chains of class II MHC protein, wherein said complex is used to detect T cells to which said complex is bound (claims 4-5, and column 6, last two lines bridging colulmn 7, first two paragraph).

It would have been obvious to one of ordinary skill in the art at the time the invention was made to biotinylate and label the soluble TCR without interchain disulfide bond, taught by the combination of the teaching of Chang et al, Gregoire et al, Garboczi et al, and Wulfing et, using the method taught by US 5,635,363, to detect said soluble TCR.

Moreover, it would have been obvious to biotinylate said soluble TCR at the C-terminus, because one would have expected that the C-terminus is not the site where the alpha and beta chains are, which contain the recognition site for antigen necessary for the action of the TCR, and thus biotinylation at the C-terminus would be least likely to interfere with antigen binding, as compared to biotinylation at the N-terminus.

One of ordinary skill in the art is motivated to biotinylate and label said soluble TCR with a reasonable expectation of success.

**REFERENCES WITH RELEVANT INFORMATION**

**1)** Weber, S et al, 1992, Nature, 356 (6372): 793-6, IDS of 03/25/02 teach that the soluble chimeric TCRs comprising alpha- and beta- chains linked to the immunoglobulin light chain constant region (Ck) is secreted at about 10 ug/ml (p.794, second column, second paragraph).

**2)** Wulfing C et al, 1995, The Immunologist, 3(2): 59-66, IDS of 03/25/02, teach that the yield of chimeric Ck fusion of both chains of TCRs is 10 mg/ml in medium (table 3 on page 63, item from ref 27).

Any inquiry concerning this communication or earlier communications from the examiner should be directed to MINH-TAM DAVIS whose telephone number is 571-272-0830. The examiner can normally be reached on 9:30AM-4:00PM.

If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, YVONNE EYLER can be reached on 571-272-0871. The fax phone number for the organization where this application or proceeding is assigned is 703-872-9306.

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MINH TAM DAVIS

PATENT EXAMINER

February 04, 2004